November 2003

DIETARY TOXICITY TEST FOR 2% DRC-1339-TREATED BROWN RICE ON NONTARGET AVIAN SPECIES

John L. Cummings
USDA-APHIS-Wildlife Services

Darryl L. York
USDA-APHIS-Wildlife Services

Kirk J. Shively
USDA-APHIS-Wildlife Services

Patricia A. Pipas
USDA-APHIS-Wildlife Services

Randal S. Stahl
USDA-APHIS-Wildlife Services, randal.s.stahl@aphis.usda.gov

See next page for additional authors

DIETARY TOXICITY TEST FOR 2% DRC-1339-TREATED BROWN RICE ON NONTARGET AVIAN SPECIES

JOHN L. CUMMINGS, United States Department of Agriculture, National Wildlife Research Center, 4101 LaPorte Avenue, Fort Collins, CO 80525, USA

DARRYL L. YORK, United States Department of Agriculture, National Wildlife Research Center, 4101 LaPorte Avenue, Fort Collins, CO 80525, USA

KIRK J. SHIVELY, United States Department of Agriculture, National Wildlife Research Center, 4101 LaPorte Avenue, Fort Collins, CO 80525, USA

PATRICIA A. PIPAS, United States Department of Agriculture, National Wildlife Research Center, 4101 LaPorte Avenue, Fort Collins, CO 80525, USA

RANDAL S. STAHL, United States Department of Agriculture, National Wildlife Research Center, 4101 LaPorte Avenue, Fort Collins, CO 80525, USA

JAMES E. DAVIS, JR., United States Department of Agriculture, National Wildlife Research Center, 4101 LaPorte Avenue, Fort Collins, CO 80525, USA

Abstract: In Louisiana and Texas DRC-1339-treated brown rice is used to manage blackbird populations that cause severe damage to newly planted rice. Nontarget bird species have been observed on some DRC-1339 bait sites. We conducted dietary toxicity tests to provide additional data on the toxicity of DRC-1339 to the following nontarget species observed on DRC-1339 bait sites: savannah sparrows (*Passerculus sandwichensis*), Canada geese (*Branta canadensis*), snow geese (*Chen caerulescens*), mourning doves (*Zenaida macroura*), western meadowlarks (*Sturnella neglecta*), and American tree sparrows (*Spizella arborea*). During our 5-day DRC-1339 dietary test, in which birds were fed only 2% DRC-1339-treated brown rice diluted 1:25 with untreated brown rice, no mortality occurred to Canada geese or snow geese, and 1 mortality occurred to savannah sparrows. Nine of 10 mourning doves, 8 of 9 meadowlarks and 8 of 10 American tree sparrows died during the same test. Most test species exhibited some degree of aversion to the DRC-1339 bait, indicating that in free-feeding situations such as DRC-1339 bait sites most nontarget birds might ingest sublethal doses and subsequently avoid treated baits. Rapid degradation and discoloration of DRC-1339 baits reduce the risk for nontarget species at DRC-1339 bait sites.

Key words: *Agelaius phoeniceus*, blackbird, DRC-1339, red-winged blackbird, rice, toxicant.

Several species of blackbirds, particularly red-winged blackbirds (*Agelaius phoeniceus*), common grackles (*Quiscalus quiscula*), boat-tailed grackles (*Quiscalus major*), great-tailed grackles (*Quiscalus mexicanus*), and brown-headed cowbirds (*Molothrus ater*) congregate in large spring roosts in Louisiana and Texas and cause extensive damage to newly planted rice. Losses to rice growers in the United States have been estimated at up to $11.5 million (Besser 1985). Blackbird damage to newly planted rice in Texas is estimated at $4.2 million annually (Decker and Avery 1990). Damage is not uniformly distributed among growers but is localized and proportional to the size of nearby blackbird roost sites (Wilson 1985). In Louisiana, damage to newly planted rice can be locally severe, with some growers reporting 100% losses (Wilson 1985).

Currently available techniques such as mechanical and pyrotechnic scaring devices, shooting, and hazing can be ineffective, costly and logistically impractical for alleviating blackbird damage (Dolbeer et al. 1994). In Louisiana and Texas, DRC-1339-treated brown rice has been widely used to reduce blackbird populations causing damage to newly planted rice. DRC-1339 avicide (3-chloro-p-toluidine hydrochloride, 3-chloro-4-methylaniline hydrochloride, or CPTH) is a slow-acting toxicant that is very highly toxic (1-10 mg/kg) to most pest birds (*Icteridae*), but moderately-to-slightly toxic (51-2,000 mg/kg) to raptors and most mammalian species (Knittle et al. 1980). The primary mode of action

of DRC-1339 is irreversible necrosis of the kidney and a subsequent inability to excrete uric acid (DeCino et al. 1966). Death occurs from uremia 24-72 h after ingestion of a lethal dose (Palmore 1978, Cunningham et al. 1979).

The use of DRC-1339-treated brown rice is restricted to a period between February 15 and March 15 in Louisiana, and between December and March in Texas. Application sites include stubble fields, bare-ground/non-crop areas and open grass sites. Although nontarget birds have been observed foraging on DRC-1339 bait sites, few mortalities have been reported (Knittle et al. 1980, Glahn et al. 1990, Cummings et al. 2002). In addition, no mortality was observed among captive savannah sparrows (*Passerculus sandwichensis*), white-crowned sparrows (*Zonotrichia leucophrys*), field sparrows (*Spizella pusilla*), song sparrows (*Melospiza melodia*), and chipping sparrows (*Spizella passerina*) fed only 2% DRC-1339-treated brown rice diluted 1:27 with untreated brown rice for 5 days (Cummings et al. 2002). A recent study of 81 DRC-1339 bait sites in Louisiana and Texas indicated frequent use by 7 nontarget species (Pipas et al. 2003). The purpose of our study is to provide additional data on the dietary toxicity of DRC-1339-treated brown rice to some of these species: savannah sparrows (*Passerculus sandwichensis*), Canada geese (*Branta canadensis*), snow geese (*Chen caerulescens*), mourning doves (*Zenaida macroura*), western meadowlarks (*Sturnella neglecta*), and American tree sparrows (*Spizella arborea*).

METHODS

We conducted a 5-day dietary toxicity test for DRC-1339-treated brown rice fed to savannah sparrows, Canada geese, snow geese, western meadowlarks, mourning doves, and American tree sparrows following the U. S. Environmental Protection Agency (USEPA) guidelines modified for 1 dose level. The Pocatello Supply Depot, Pocatello, Idaho, formulated and supplied the 2% DRC-1339-treated brown rice. The National Wildlife Research Center verified its purity and percent formulation. The DRC-1339-treated brown rice is diluted 1:25 with untreated brown rice for testing.

Following capture, all birds were weighed, banded and placed in individual pens for a 14-day quarantine and acclimation period. Perches were provided in each pen and birds were allowed free access to food (a combination of brown rice, small grains, and Purina game bird starter) and water. We divided each species into 2 groups, control (n=10) and treated (n=10) for testing. During the last 3 days of acclimation, birds received brown rice only. We weighed each test bird at the end of the acclimation period. The following morning at 0730 h test birds were offered from 50 to 300 g of feed depending on the species. The control group was offered untreated brown rice only and the treated group was offered 2% DRC-1339-treated brown rice diluted 1:25 with untreated brown rice. Food was placed in a standard plastic dish appropriate for the species being tested and placed on an oversized pan (50 x 50 cm) to collect spillage. At the same time, 3 food dishes containing the species appropriate amount of control and treated food, respectively, were placed at a random location outside the pen to measure any effects from moisture. We collected and weighed the food from each pen and the moisture samples daily at 1530 h. Each test day, birds were offered a fresh sample of assigned food. We repeated the entire process for 5 consecutive days.

We checked birds daily for signs of DRC-1339 poisoning and noted the time birds were incapacitated (sedentary) or dead. Dead birds were collected, weighed and examined internally for white deposits of uric acid in the pericardial cavity which is a common sign of DRC-1339 poisoning (DeCino et al. 1966). In addition, we removed the GI tract and breast tissue from each dead bird for DRC-1339 residue analysis. Birds from each group surviving the test were monitored from 3 to 5 days post treatment. Following this period, we euthanized the remaining birds and randomly selected 3 birds of each species from each treatment group for DRC-1339 residue analysis. DRC-1339 residues in the GI tract and breast tissues were analyzed using the method of Stahl et al. (2002). Birds were assumed to have no detectable DRC-1339 residues in the tissues when the level observed was less than the method limit of quantification (LOQ). The LOQ was defined as signal peak height for a DRC-1339 fortified sample of known concentration that is 10 times the average baseline noise for a nonfortified control sample when measured from the top to the bottom of the peaks. This was the lowest level that can be quantified from a linear regression of peak height versus fortification concentration.

Descriptive statistics were used to compare the mortality among treatment groups of each species.

RESULTS

Savannah Sparrows

There was 1 mortality in the DRC-1339 group and none in the control group (Table 1). Daily food consumption by savannah sparrows in the DRC-1339 group was about 30% more than savannah sparrows in the control group. A necropsy of 3 savannah sparrows from each treatment group and 1 savannah sparrow that died during testing showed no signs of DRC-1339 poisoning. DRC-1339 residue analysis of the gastrointestinal tract and breast tissue from each of these birds indicated no DRC-1339 residues (Table 2).

Table 1. Dietary toxicity test of 2% DRC-1339-treated brown rice to nontarget birds, Fort Collins, Colorado, 2000-2002.

Species	Sample n	Treatment	Weight (Pre) ($\bar{x}$ g)	Weight (Post) ($\bar{x}$ g)	Daily consumption ($\bar{x}$ g/bird)	Survived
Savannah sparrow	10	Control	14.6	13.6	2.0	10
Savannah sparrow	10	DRC-1339	14.8	13.3	2.6	9
Canada goose	10	Control	3,970	4,070	148.9	10
Canada goose	10	DRC-1339	3,720	3,760	18.4	10
Snow goose	10	Control	2,260	1,477	9.9	10
Snow goose	10	DRC-1339	2,210	1,554	3.0	10
Western meadowlark	10	Control	91.8	82.8	7.0	10
Western meadowlark	9	DRC-1339	90.6	77.9[b]	2.6[a]	1
Mourning dove	10	Control	108.2	108.2	6.9	10
Mourning dove	10	DRC-1339	114.4	99.9[b]	0.2[a]	1
American tree sparrow	10	Control	18.3	Unknown	5.6	10
American tree sparrow	10	DRC-1339	18.1	Unknown	3.1	2

[a]Consumption of remaining birds.

[b]Includes weight of birds at death.

Canada Geese

There was no mortality of Canada geese in the DRC-1339 group or control group (Table 1). Daily food consumption of Canada geese in the DRC-1339 group averaged about 18.4 g per bird, whereas birds in the control group averaged 148.9 g/bird. Canada geese developed an aversion to DRC-1339-treated brown rice following their initial exposure. Their aversion to DRC-1339-treated brown rice did not cause any significant weight loss. A necropsy of 3 Canada geese from each treatment group showed no signs of DRC-1339 poisoning. DRC-1339 residue analysis of the gastrointestinal tract and breast tissues from each of these birds indicated that there were no DRC-1339 residues (Table 2).

Table 2. DRC-1339 residues in the gastrointestinal tract and breast tissue of nontarget birds fed 2 % DRC-1339-treated brown rice during a 5-day dietary toxicity test, Fort Collins, Colorado, 2000-2002.

Species	Sample n	Treatment	GI tract $\bar{x}$	(ug/g) Range	Breast tissue $\bar{x}$	(ug/g) Range	LOQ[a] (ug/g) GI tract	LOQ (ug/g) Breast tissues
Savannah sparrow	3	Control	<LOQ	<LOQ	<LOQ	<LOQ	0.053	0.053
Savannah sparrow	3	DRC-1339	<LOQ	<LOQ	<LOQ	<LOQ	0.053	0.053
Canada goose	3	Control	<LOQ	<LOQ	<LOQ	<LOQ	0.163	0.05
Canada goose	3	DRC-1339	<LOQ	<LOQ	<LOQ	<LOQ	0.163	0.05
Snow goose	3	Control	<LOQ	<LOQ	<LOQ	<LOQ	0.072	0.059
Snow goose	3	DRC-1339	<LOQ	<LOQ	<LOQ	<LOQ	0.072	0.059
Western meadowlark	3	Control	<LOQ	<LOQ	<LOQ	<LOQ	0.157	0.031
Western meadowlark	3	DRC-1339	1.48	0.85-2.5	0.27	0.2-0.37	0.157	0.031
Mourning dove	10	Control	<LOQ	<LOQ	<LOQ	<LOQ	0.053	0.178
Mourning dove	10	DRC-1339	0.17	<LOQ-1.4	<LOQ	<LOQ	0.053	0.178
American tree sparrow	3	Control	<LOQ	<LOQ	<LOQ	<LOQ	0.084	0.04
American tree sparrow	10	DRC-1339	1.04	<LOQ-3.8	0.102	<LOQ-0.23	0.084	0.04

[a]Limit of quantification

Snow Geese

There was no mortality of snow geese in the DRC-1339 group or control group (Table 1). Overall, daily food consumption by snow geese averaged only 10 g/bird for the control group and 3 g/bird for the DRC-1339 group. Both groups lost more than 30% of their body weight during the test period. A necropsy of 3 snow geese from each treatment group showed no signs of DRC-1339 poisoning. DRC-1339 residue analysis of the tract and breast tissues from each of these birds indicated no DRC-1339 residues (Table 2).

Western Meadowlark

In the DRC-1339 group, 8 of 9 western meadowlarks died during testing (Table 1). All meadowlarks in the control group survived. One meadowlark in the DRC-1339 group died in (24 h; 6 meadowlarks died between 24 to 48 h; and 1 meadowlark died 120 h after the start of the test. The daily food consumption of the surviving bird averaged 4.2 g. Meadowlarks in the DRC-1339 group exhibited an aversion to the DRC-1339 bait mix during the test. Overall, daily food consumption of these birds averaged 2.6 g, whereas the daily food consumption for meadowlarks in the control group averaged 7.0 g. A necropsy of all meadowlarks from the DRC-1339 group showed that seven birds had white deposits of uric acid in the pericardial cavity. The surviving bird showed no signs of DRC-1339 poisoning. DRC-1339 residues for meadowlarks in the DRC-1339 group averaged 1.48 ug/g in the gastrointestinal tract and 0.272 ug/g in breast tissue (Table 2). A necropsy of 3 meadowlarks in the control groups showed no signs of DRC-1339 poisoning or DRC-1339 residues in the gastrointestinal tract or breast tissue.

Mourning Doves

In the DRC-1339 group, 9 of 10 mourning doves died during the test (Table 1). All mourning doves in the control group survived. Four mourning doves in the DRC-1339 group died between 24 to 48 h; 2 mourning doves died between 48 to 72 h; and 3 mourning doves died between 72 to 96 h after the start of the test. Mourning doves in the DRC-1339 groups exhibited an aversion to the DRC-1339 bait mix during the test. The daily food consumption of these birds averaged about 0.2 g/bird, whereas birds in the control group averaged 7 g/bird. Most food consumption in the DRC-1339 group took place on day 1 of the test where mourning doves consumption averaged 0.7 g/bird. The surviving bird in the DRC-1339 group consumed 0.78 g of DRC-1339 food on day 1 and 0.1 g of DRC-1339 food over the remaining 4 days of the test. During the test period, this bird weight dropped from 120 g to 102 g. A necropsy of the 10 mourning doves from the DRC-1339 group, including the surviving bird showed that 8 of 10 had white deposits of uric acid in the pericardial cavity. The surviving bird from this group showed no signs of DRC-1339 poisoning. DRC-1339 residues for mourning doves in the DRC-1339 group averaged 0.17 ug/g of DRC-1339 in the gastrointestinal tract and were LOQ in breast tissue (Table 2). A necropsy of 3 mourning doves in the control groups showed no signs of DRC-1339 poisoning or DRC-1339 residues in the gastrointestinal tract or breast tissue.

American Tree Sparrows

In the DRC-1339 group, 8 of 10 American tree sparrows died during the test (Table 1). All American tree sparrows in the control group survived. Five American tree sparrows died in 24 h; 1 American tree sparrow died between 24 to 48 h; and 2 American tree sparrows died between 48-72 h after the start of the test. American tree sparrows in the DRC-1339 group exhibited an aversion to DRC-1339 during the test. The daily food consumption for these birds averaged 3.1 g/bird where as birds in the control group averaged 5.6 g/bird. A necropsy of 10 American tree sparrows from the DRC-1339 group including the 2 surviving birds showed 8 of 10 had white deposits of uric acid in the pericardial cavity. The surviving birds from this group showed no signs of DRC-1339 poisoning. DRC-1339 residues for American tree sparrows in the DRC-1339 group averaged 1.04 ug/g of DRC-1339 in the gastrointestinal tract and 0.102 ug/g DRC-1339 in breast tissue (Table 2). A necropsy of 3 American tree sparrows in the control groups showed no signs of DRC-1339 poisoning or DRC-1339 residues in the gastrointestinal tract or breast tissue.

DISCUSSION

In Louisiana and Texas DRC-1339-treated brown rice is a USEPA-registered product that is used to manage blackbird populations that cause severe damage to newly planted rice. Several studies of DRC-1339 hazards to nontarget birds have reported no or few mortalities (Knittle et al. 1980, Glahn et al. 1990, Cummings et al. 2002). In addition, no mortality was observed when nontarget species (savannah sparrows, white-crowned sparrows, field sparrows, song sparrows, and chipping sparrows) were captured from around DRC-1339 bait sites, held in captivity and fed only 2% DRC-1339-treated brown rice diluted 1:27 with untreated brown rice for 1 and 12 h each day for 5 days (Cummings et al. 2002). Monitoring studies of nontarget use of 81 DRC-1339 bait sites in Louisiana and Texas between 2000 and 2002, identified primarily 7 species, savannah sparrows, snow geese, killdeer (*Charadrius vociferus*), American pipits (*Anthus rubescens*), northern cardinals (*Cardinalis cardinalis*), mourning doves, and eastern meadowlarks that were observed on bait sites (Pipas et al. 2002). Our

5-day DRC-1339 dietary test, in which birds were fed only 2% DRC-1339-treated brown rice diluted 1:25 with untreated brown rice, indicated that mourning doves and meadowlarks may be at risk. We suspect northern cardinals could also be at risk at bait sites that are close to preferred cardinal habitat. While we do not have toxicity data on American pipits or killdeer, we believe that DRC-1339 baiting poses little hazard to these species because their diet is mostly animal matter (Jackson and Jackson 2000, Verbeek and Hendricks 1994).

The 5-day dietary feeding test was designed to test the worst case scenario for exposure of nontarget birds to DRC-1339-treated brown rice formulated according to label directions. However, in reality this case would not occur in any DRC-1339 baiting program and over estimates the field mortality occurring with these species. The baiting program follows the EPA-approved label instructions for DRC-1339 baiting which has built in procedures to reduce nontarget exposure. The low numbers of nontarget species on DRC-1339 bait site can be attributed to location of bait sites, feeding activity of blackbirds, bait availability, and bait type. Bird observations of DRC-1339 bait sites showed that blackbirds can overwhelm a site and exclude nontarget birds and that most DRC-1339 baits are consumed by target birds within the first few hours of exposure (Glahn et al. 1990, Cummings et al. 1992).

Our study showed that most of the nontarget test species evaluated exhibited some degree of aversion to the toxic bait, which resulted in reduced food consumption. In free-feeding situations such as at DRC-1339 bait sites in Louisiana or Texas, nontarget birds would be more apt to ingest sublethal doses or avoid treated baits. Additionally, DRC-1339 degrades rapidly under environmental conditions, losing 55% of its potency within 3 days (Primus et al. 1997). DRC-1339 baits turn brown within 24 h of exposure, which might repel many birds from ingesting treated baits. All of these factors combine to reduce the risk for nontarget species that use DRC-1339 bait sites.

ACKNOWLEDGMENTS

We thank the Louisiana Rice Research Board, Louisiana Rice Growers Association, Louisiana State University Rice Research Station, and USA Rice Foundation for support. We also thank Mark Tobin, George Linz and John Eisemann for their reviews.

LITERATURE CITED

Besser, J. F. 1985. A grower's guide to reducing bird damage to U.S. agricultural crops. Denver Wildlife Research Center, Bird Section Research Report No. 340.

Cummings, J. L., P. A. Pochop, R. M. Engeman, J. E. Davis, and T. M. Primus. 2002. Evaluation of Flight Control® to reduce blackbird damage to newly planted rice in Louisiana. International Biodeterioration and Biodegradation 49:169-173.

Cummings, J. L., J. E. Glahn, E. A. Wilson, J. E. Davis, jr., D. L. Bergman, and G. A. Harper. 1992. Efficacy and non-target hazards of DRC-1339 treated rice baits used to reduce roosting populations of depredating blackbirds in Louisiana. National Wildlife Research Center, Bird Section Research Report 481.

Cunningham, D. J., E. W. Schafer, jr., and L. K. McConnell. 1979. DRC-1339 and DRC-2698 residues in starlings: preliminary evaluation of their secondary hazard potential. Proceedings of the Bird Control Seminar 8:31-37

Decino, T. J., D. J. Cunningham, and E. W. Schafer, jr. 1966. Toxicity of DRC-1339 to starlings. Journal of Wildlife Management 30:249-253.

Decker, D. G., and M. L. Avery. 1990. Reducing blackbird damage to newly planted rice with a nontoxic clay-based seed coating. Proceedings of the Vertebrate Pest Conference 14:327-331.

Dolbeer, R. A., N. R. Holler, and D. W. Hawthorne. 1994. Identification and control of wildlife damage. Pages 474-506 *in* T. A. Bookhout, editor. Research and management techniques for wildlife and habitats. The Wildlife Society, Bethesda, Maryland, USA.

Glahn, J. F., E. A. Wilson, and M. L. Avery. 1990. Evaluation of a DRC-1339 baiting program to reduce sprouting rice damage caused by spring roosting blackbirds. National Wildlife Research Center, Bird Section Research Report 448.

Jackson, B. J. S., and J. A. Jackson. 2000. Killdeer (*Charadrius vociferous*). The birds of North America No. 517. The American Ornithologists' Union, Washington, D. C., USA and The Academy of Natural Sciences, Philadelphia, Pennsylvania, USA.

Knittle, C. E., J. L. Guarino, J. C. Nelson, R. W. DeHaven, and D. J. Twedt. 1980. Baiting blackbirds and starling congregating areas in Kentucky and Tennessee. Proceedings of the Vertebrate Pest Conference 9:31-37.

Palmore, W. P. 1978. Diagnosis of toxic acute renal failure in cats. Florida Veterinary Journal 7: 14-15, 36-37.

Pipas, P. A., J. L. Cummings, J. D. Eisemann, and R. M. Engeman. 2003. Nontarget bird use of DRC-1339 bait sites during operational baiting programs in Louisiana and Texas. *In* G. M. Linz, editor. Management of North American blackbirds. National

Wildlife Research Center, Fort Collins, Colorado, USA. In Press.

Primus, T. M., J. N. Tawara, J. J. Johnston, J. L. Cummings, S. A. Volz, M. J. Goodall, D. B. Hurlbut, D. L. Griffin, and S. Turnipseed. 1997. Identification of degradation products of the avicide 3-chloro-p-toluidine hydrochloride in Louisiana rice fields. Environmental Science and Technology 31:346-350.

Stahl, R. S., T. W. Custer, P. A. Pochop, and J. J. Johnston. 2002. Improved method for quantifying the avicide 3-chloro-p-toluidine hydrochlorine in bird tissues using a deuterated surrogate/gc/ms method. Journal of Agricultural and Food Chemistry 50:732-738.

Verbeek, N. A. M., and P. Hendricks. 1994. American pipit (*Anthus rubescens*). The birds of North America No. 95. The Birds of North America, Philadelphia, Pennsylvania, USA.

Wilson, E. A. 1985. Blackbirds depredation on rice in southwestern Louisiana. Thesis, Louisiana State University, Baton Rouge, USA.

2003. G. M. Linz, editor. Management of North American Blackbirds (Proceedings of a special symposium of the Wildlife Society 9th Annual Conference. Bismarck, North Dakota, September 27, 2002).